Interference of the polymeric material of swabs with the quantification of extracellular polymeric substances in biofilm samples

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INTRODUCTION

The detection of biofilms is an important challenge in the food industry. Biofilms are microorganisms that are attached to a surface and live in a self-produced matrix of extracellular polymeric substances (EPS) containing among others carbohydrates, uronic acids and proteins. They may be a source of contamination of food products with pathogens, spoilage organisms and spoilage enzymes. Most existing biofilm detection methods only focus on microorganisms. As a biofilm consists of both microorganisms and EPS, it is necessary to measure the EPS components as well. However, it may be possible that the polymeric material of the swabs used for the biofilm sampling from the food contact surfaces contain and release sugars, uronic acids or proteins during sample preparation. Therefore the possible interference of different swab-types with the detection of EPS was investigated in this study.

MATERIAL & METHODS

Flock swab (nylon fibers)

Sponge stick (cellulose)

+ 10 mL physiological saline

This is to keep microbial cells in a biofilm sample intact without interference with the chemical determinations (↔ broths).

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stomacher 1’

This is to release a biofilm sample from the swab.

↓

sonicate 3 x 30” at 50% amp. and 0,5 cycle

This is to separate the EPS from bacterial cells in biofilm samples.

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centrifuge 10 min at 13000 g

supernatant containing EPS and possibly dissolved chemical constituents of the swab

pellet containing bacterial cells and possibly solid particles released from the swab

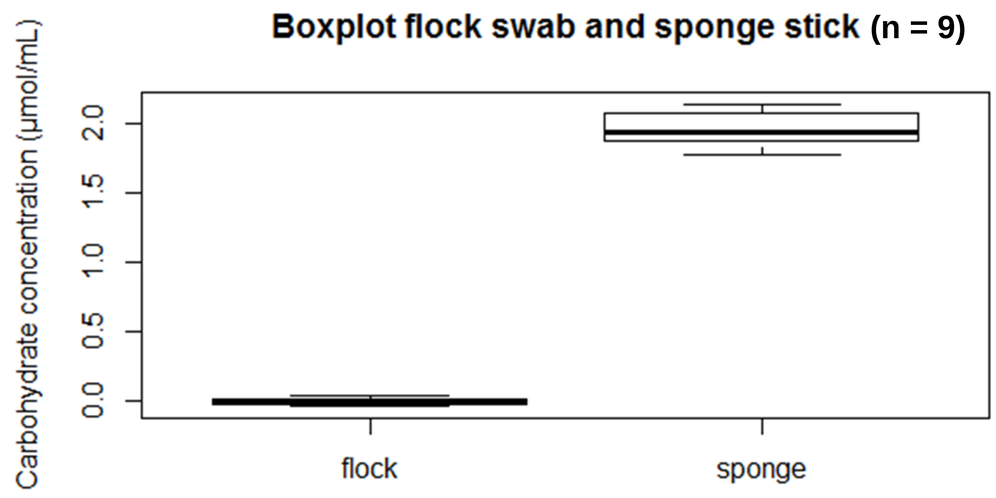
Carbohydrate content by phenol-sulfuric acid method measured at 492 nm using glucose as a standard

Uronic acid content by a method using m-hydroxydiphenyl and sulfuric acid measured at 540 nm using D-galacturonic acid as a standard

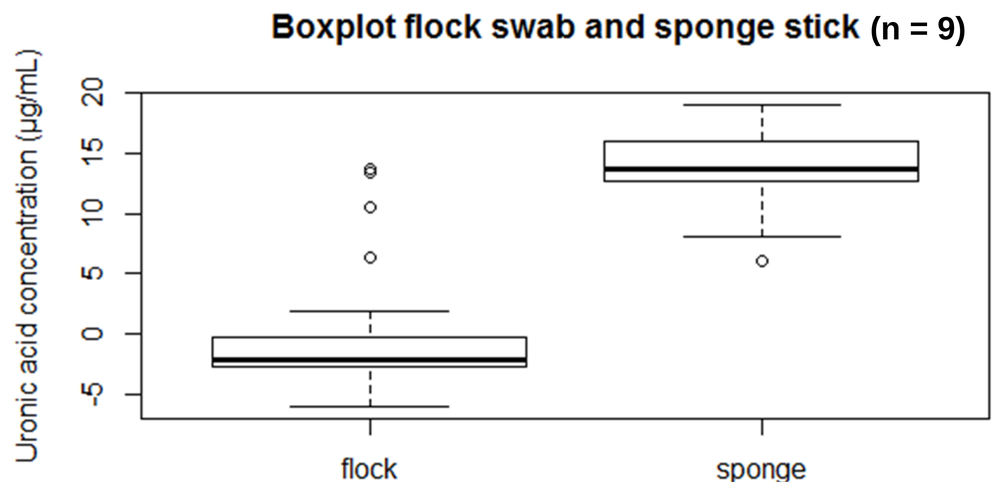
Protein content by the Bradford method measured at 595 nm using bovine serum albumin as a standard

RESULTS

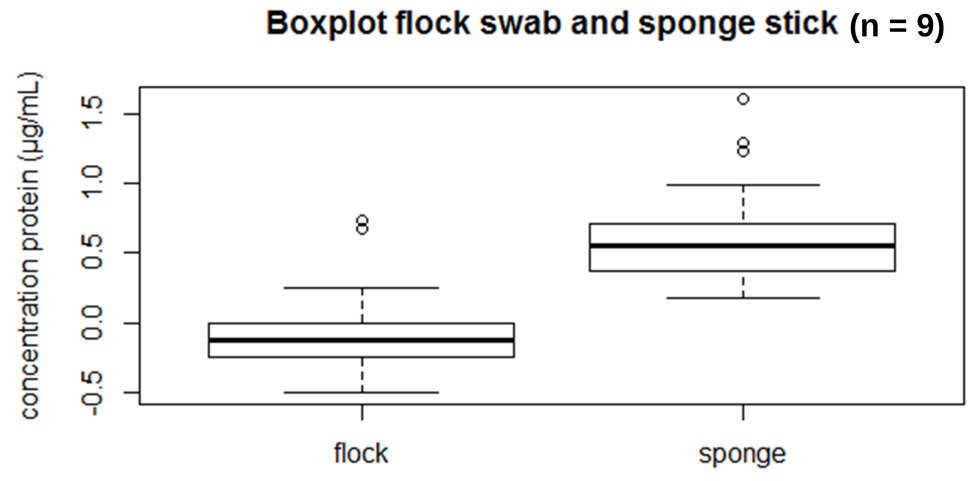
CARBOHYDRATES



URONIC ACIDS



PROTEINS



CONCLUSION

Results indicated a need to test the swabs to be used for sampling first as interference with the EPS analysis was found. The distribution of the concentrations found in the swabs can be used to calculate an adjusted limit of detection (LOD) for the carbohydrate, uronic acid and protein content of EPS in biofilm samples. Among the tested swabs, the flock swabs are best suited for biofilm sampling as the cellulose sponge stick released biopolymers that interfere with the EPS analysis.

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